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TITLE: 99-Technetium Sestamibi Scanning to Predict the Efficacy of Estramustine Phosphate in Overcoming Paclitaxel Resistance in Patients with Advanced Breast Cancer

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13. ABSTRACT *(Maximum 200 Words)*

The purpose of this research is to investigate the ability of 99-Technetium Sestamibi (Tc-99-SM) to serve as a non-invasive means of assessing the presence of clinically relevant drug resistance in patients with advanced breast cancer. Tc-99-SM is a substrate of p-glycoprotein, the transmembrane drug efflux transporter involved in classic multi-drug resistance (MDR). Our hypothesis is that rapid clearance of Tc-99-SM correlates with the presence of functional multi-drug resistance and can be used to predict which patients will have tumors resistant to chemotherapy drugs that are MDR substrates. We have demonstrated marked variability in the tumor clearance of Tc-99-SM among patients. More importantly, we have initiated a clinical protocol that will investigate whether changes in the clearance of 99-Tc-SM observed before and after the administration of an MDR inhibitor, can predict how effective the inhibitor will be in overcoming clinical drug resistance in individual patients.

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Introduction

The purpose of this research is to investigate the ability of 99-Technetium Sestamibi (Tc-99-SM) to serve as a non-invasive means of assessing the presence of clinically relevant drug resistance in patients with advanced breast cancer. Tc-99-SM is a substrate of the p-glycoprotein, the transmembrane drug efflux transporter involved in classic multi-drug resistance (MDR). We are testing the hypothesis is that rapid clearance of Tc-99-SM correlates with the presence of functional MDR and can be used to predict which patients will have tumors resistant to chemotherapy drugs that are MDR substrates. More importantly, we are investigating whether changes in the tumor clearance of 99-Tc-SM observed before and after the administration of an MDR inhibitor, can predict whether the inhibitor can overcome clinical drug resistance in an individual patient.

Body

Task 1: *Complete a clinical trial of estramustine/paclitaxel in patients with advanced cancer of the breast refractory to paclitaxel, Months 1-30:*

- *Finalize clinical protocol. Obtain Institutional Review Board approval*
- *Recruit patients from the clinics of Bellevue and Tisch Hospital who have advanced breast cancer and are candidates for treatment with paclitaxel. Initiate treatment with paclitaxel.*
- *At the time each enrolled patient demonstrates resistance to paclitaxel, begin estramustine/paclitaxel. Patients may demonstrate primary resistance to paclitaxel (no response to an adequate trial of paclitaxel) or secondary resistance (failure following an initial response to paclitaxel).*

We have made progress in implementing Task 1. We have finalized the clinical treatment protocol required by this research project. During the process of finalizing the research protocol, new information about the interaction between the estramustine and p-glycoprotein became available. Specifically, a study of the pharmacokinetics of paclitaxel given concurrently with estramustine indicates that estramustine does not inhibit p-glycoprotein or otherwise affect drug efflux from tumor cells.[1] While this clinical finding is at odds with prior laboratory studies indicating an inhibitory effect of estramustine on drug efflux[2, 3], it strongly casts doubt on the ability of estramustine to serve as a clinical inhibitor of MDR. We therefore investigated the use of other agents that are more likely to successfully inhibit drug efflux and decided to replace estramustine with the biricodar dictrate (VX-710, Incel™) as the MDR inhibitor for purposes of this study. We chose biricodar because of recent studies demonstrating that it is a potent inhibitor of both p-gp and the multi-drug resistance associated protein (MRP) at concentrations well tolerated in human studies.[4, 5] A phase I trial of biricodar given as a 24-hour infusion in combination with 3-hour paclitaxel established that the drugs may be given safely together under conditions that simulate optimal MDR

inhibition in vitro. In combination with 24-hour biricodar, paclitaxel at a dose of 80 mg/m^2 achieves exposure comparable to the standard 3 hour 175 mg/m^2 paclitxel (similar AUC, Vss, plasma concentration time above 0.05 microM), however, paclitaxel clearance is reduced by approximately 50%.[5] This has been confirmed in a phase II study of biricodar and paclitaxel in patients with taxane-refractory breast cancer. Initial results from this study are promising, demonstrating partial responses in 4 of 35 (11.4%) evaluable patients (unpublished data, Vertex pharmaceuticals). In the phase II setting, the toxicity of the combined regimen, biricodar 120 mg/m^2/hr x 24 hours and paclitaxel 80 mg/m^2 infused over 3 hours, was modest and resembled the toxicity of paclitaxel given as a single agent via a 24 hour infusion. Our protocol has been approved by the Institutional Review Board at New York University and has been given preliminary approval by the Surgeon General's Human Research Review Board. We expect to begin enrolling patients within the next four to six weeks.

Task 2: *Concurrently with Task 1, complete an imaging study evaluating serial tc-99-SM scanning to assess the presence of functional drug efflux at three critical time points in the treatment of patients during the clinical trial described in Task 1, Months 1- 30:*

- *Baseline Tc-99-SM scans will be performed before the administration of therapy with paclitaxel.*
- *At the time each patient exhibits resistance to paclitaxel, before the administration of estramustine, a second Tc-99-SM scan will be obtained.*
- *Following the administration of the first 3-day treatment with estramustine, a third Tc-99-SM scan will be obtained.*

We have made progress on Task 2. An imaging study with Tc-99-SM scanning has been approved by the Institutional Review Board at New York University and by the Surgeon General's Human Research Review Board. Under this study, we have performed Tc-99-SM scanning in 3 patients with advanced breast cancer. We have carefully analyzed the Tc-99-SM clearance data, and have found significant variability in the rate of clearance of Tc-99-SM from the patients'tumors. (See Appendix.) We believe that this represents varying degrees of expression of relevant drug efflux proteins (p-gp and/or MRP) in these patients. The next steps in our project are to determine whether biricodar, administered in the clinical trial described in Task 1, can significantly increase tumor retention of Tc-99-SM, and whether the change in retention is reflected clinically as reversal of drug resistance to paclitaxel.

Task 3: *Data analysis and report of conclusions Months 31-36:*

- *Evaluate correlations between tc-99-SM clearance, response to paclitaxel, and the efficacy of estramustine in overcoming paclitaxel resistance.*
- *A report of the conclusions and an initial manuscript will be prepared.*

Not applicable.

Key Research Accomplishments

- We have performed preliminary studies of Tc-99-SM scanning in patients with advanced breast cancer and found variability in the clearance of Tc-99-SM suggesting that altered drug efflux may be a significant mechanism of drug resistance in some patients.
- We have begun a clinical trial of paclitaxel and the drug resistance reversing agent biricodar in patients with advanced breast cancer. The trial includes correlative Tc-99-SM scanning, allowing us to test the hypothesis that a decrease in Tc-99-SM clearance from tumor after the administration of biricodar will predict for the restoration of taxane sensitivity.

Reportable Outcomes

There are not yet reportable outcomes from this work.

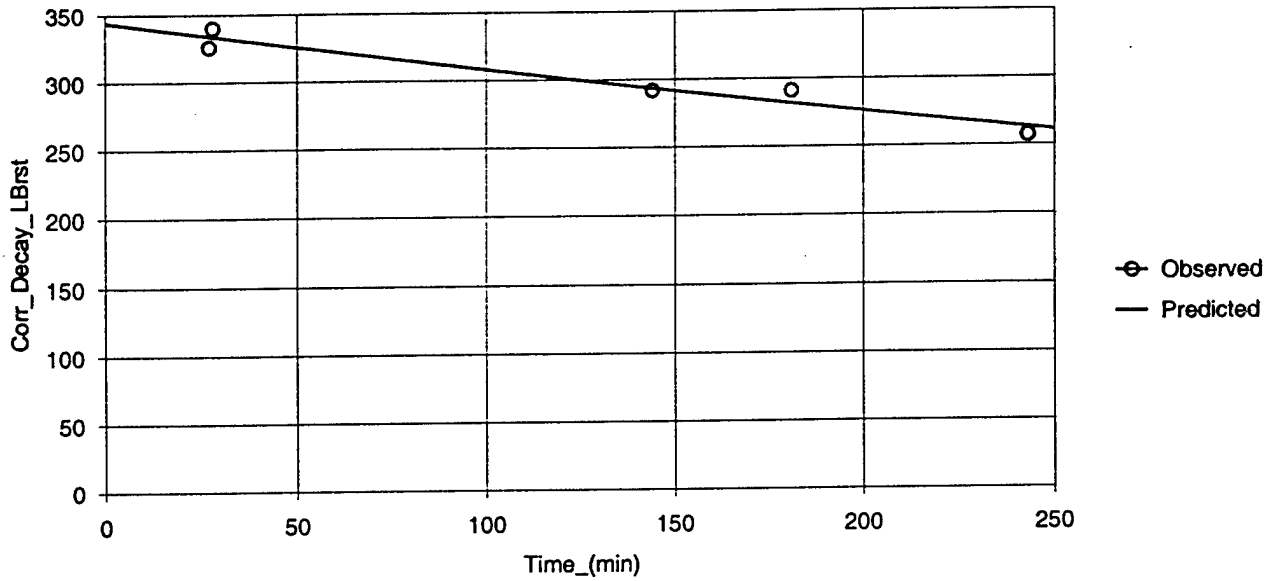
Conclusions

At present, our conclusions are limited. Consistent with our hypothesis that the rate of Tc-99-SM clearance reflects the expression of drug efflux proteins, we have observed significant intrapatient variation in the studies of tumor clearance of Tc-99-SM.

References

1. Garcia A, Keren-Rosenberg S, Parimoo D, Muggia F. Phase I and pharmacologic study of estramustine phosphate and short infusions of paclitaxel in women with solid tumors. J Clinical Oncology 1998; 16:2959-2963.
2. Speicher La, Barone Lr, Chapman Ae, et al. P-glycoprotein binding and modulation of the multidrug-resistant phenotype by estramustine. Journal of the National Cancer Institute 1994; 86:688-94.
3. Yang Cp, Shen Hj, Horwitz Sb. Modulation of the function of P-glycoprotein by estramustine. Journal of the National Cancer Institute 1994; 86:723-5.
4. Germann UA, Shlyakhter D, Mason VS, et al. Cellular and biochemical characterization of VX-710 as a chemosensitizer: reversal of P-glycoprotein-mediated multidrug resistance in vitro. Anti-Cancer Drugs 1997; 8:125-40.
5. Rowinsky EK, Smith L, Wang YM, et al. Phase I and pharmacokinetic study of paclitaxel in combination with biricodar, a novel agent that reverses multidrug resistance conferred by overexpression of both MDR1 and MRP. Journal of Clinical Oncology 1998; 16:2964-76.

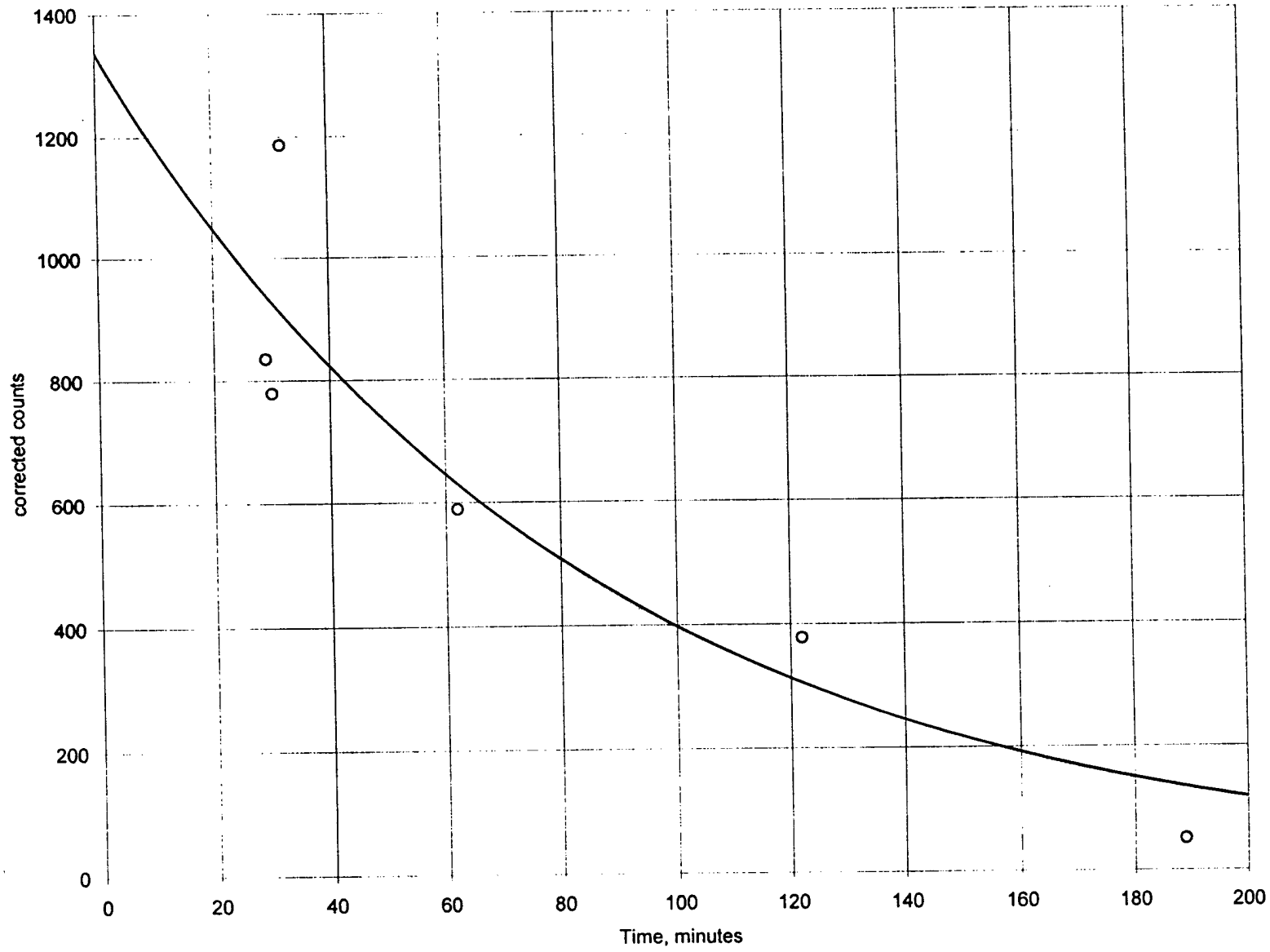
Appendices



$t\,1/2 = 620$ min.

Patient #!. Patient's tumor shows marked retention of Tc-99-SM suggesting that drug efflux mediated resistance will not occur.

Patient #2. Tumor deomstrates rapid clearance (minimal retention) of of Tc-99-SM suggesting that drug efflux from tumor may be a clinically relevant mechanism of drug resistance in this patient.



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